1. create amine-rich surface
2. attach agent-immobilizing material

3. Immobilize Different Agents

Fig. 1

Fig. 2

Fig 3

Collagen I/Fibronectin = 1   Fibroblast Growth Factor-7/Neuropeptide Y = 6
Collagen III/Vitronectin = 2   Growth Hormone/Interleukin-3 = 7
Collagen IV/Laminin = 3   Prolactin/Hepatocyte Growth Factor = 8
Collagen VI/Polylysine = 4   Interleukin-18/Neurturin = 9
Elastin/Polyornithine = 5   Cholesterol/Midkine = 10
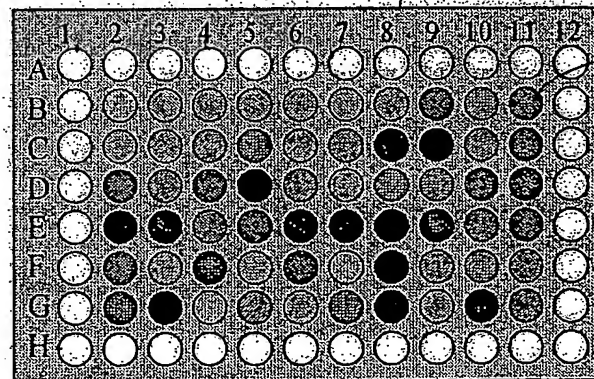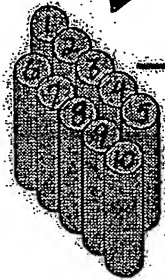
Fig. 4

Fig 5

A.

Case 1: All 10 factors are present
Overall factor concentration = $[10/10] = [1]$
$[1]$ factor/well

B.

Case 2: 5 out of 10 factors are present
Overall factor concentration = $[5/10] = [0.5]$
$[0.5]$ factor/well

Fig. 6

A.

Case 1 : All 10 factors are present

Overall factor concentration = $[^{10}/_{10}] \cdot [1]$

$[1]$ factor/well

B.

Case 2 : 5 out of 10 factors are present

Overall factor concentration = $[1]$

$[1]$ factor/well
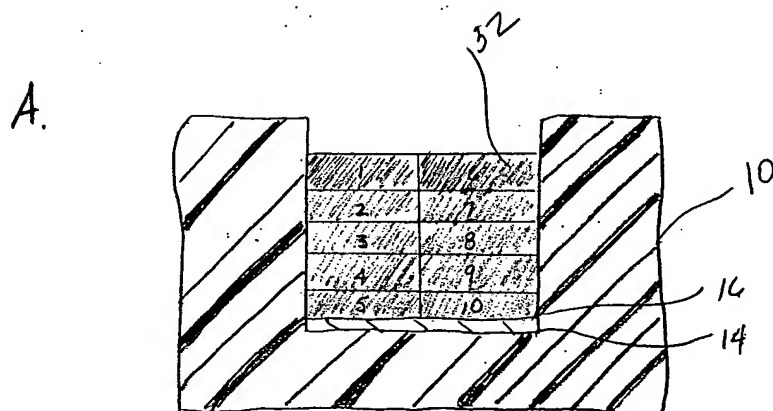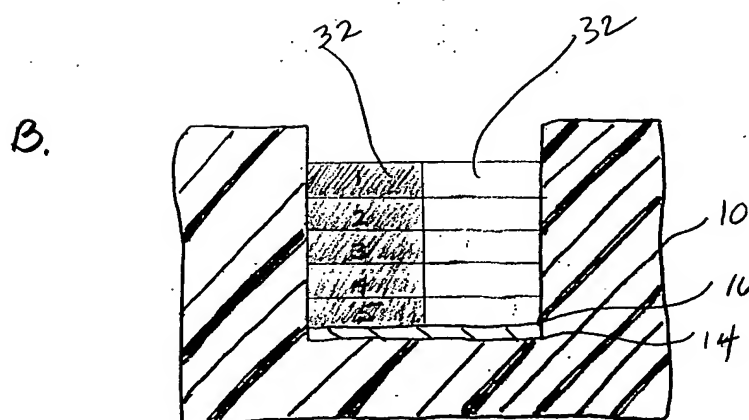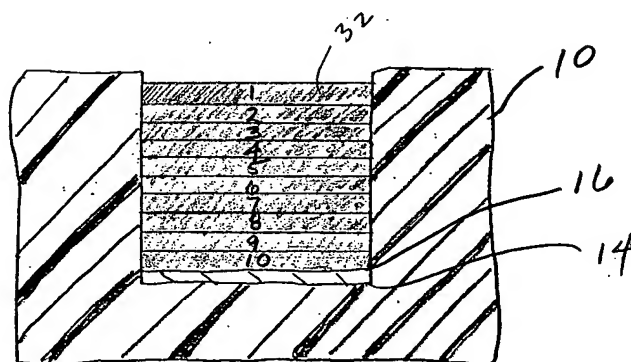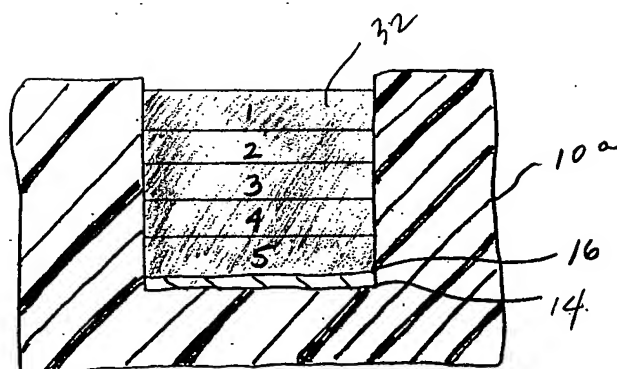
Run	Type	A:Fibronectin (µl)	B:Collagen I (µl)	C:Vitronectin (µl)	D:Collagen VI (µl)	E:Collagen III (µl)	F:Laminin (µl)	G:Collagen Iv (µl)	H:Elastin (µl)	J:Poly-L-Lysine (µl)	K:Poly-L-Ornithine (µl)
1	CentEdge			25		25					
2	CentEdge	25							25		
3	CentEdge				25				25		
4	Vertex									50	
5	CentEdge				25						25
6	CentEdge	25	25								
7	CentEdge			25						25	
8	CentEdge					25			25		
9	Vertex			50							
10	CentEdge	25						25			
11	Center	5	5	5	5	5	5		5	5	5
12	Vertex							50			
13	CentEdge	25						25			
14	CentEdge	25			25						
15	CentEdge	25									25
16	CentEdge		25				25				
17	CentEdge					25	25				
18	Center	5	5	5	5	5	5	5	5	5	5
19	Center	5	5	5	5	5	5	5	5	5	5
20	CentEdge				25		25				
21	CentEdge					25		25			
22	CentEdge		25						25		
23	CentEdge	25				25					
24	CentEdge			25	25						
25	Vertex		50								
26	Vertex	50									
27	CentEdge				25			25			
28	Vertex					50					
29	CentEdge			25						25	
30	CentEdge		25								25
31	Vertex						50				
32	Vertex						50			25	
33	CentEdge				25					25	
34	CentEdge		25			25					
35	CentEdge				25				25		
36	CentEdge					25				25	
37	CentEdge		25						25		
38	CentEdge		25								25
39	Vertex								50		
40	CentEdge				25	25					
41	Center	5	5	5	5	5	5	5	5	5	5
42	Vertex										50
43	CentEdge	25								25	
44	CentEdge	25		25							
45	CentEdge		25					25			
46	CentEdge		25	25							
47	CentEdge		25							25	
48	Vertex					50					
49	CentEdge			25				25			
50	CentEdge			25				25			
51	CentEdge					25					25
52	CentEdge		25			25					

Figure 7

		1	2	3	4	5	6	7	8	9	10	11	12
A													
B			VN / C III	FN / ELA	Col VI / ELA	▓▓▓	C VI / PO	FN / CI	VN / PL	C III / ELA	▓▓▓	FN / LAM	
C			MID▓	▓▓▓	FN / C IV	FN / C VI	FN / PO	C I / LAM	C III / LAM	MID▓	MID▓	C VI / LAM	
D			C III / C IV	C I / ELA	FN / C III	VN / C VI	▓▓▓	C VI / C IV	▓▓▓	VN / PL	C I / PO		
E			▓▓▓	C III	C VI / PL	C I / C III	C III / PL	C III / PL	VN / ELA	VN / PO	▓▓▓	C VI / C III	
F			MID▓	▓▓▓	FN / PL	FN / VN	C I / C IV	C I / VN	C I / PL	▓▓▓	VN / LAM	VN / C IV	
G			C III / PO	C I / C VI									
H													
			▓▓▓	Midpoint - contains all 10 adhesion ligands									
			▓▓▓	Single adhesion ligand containing wells									

Figure 8
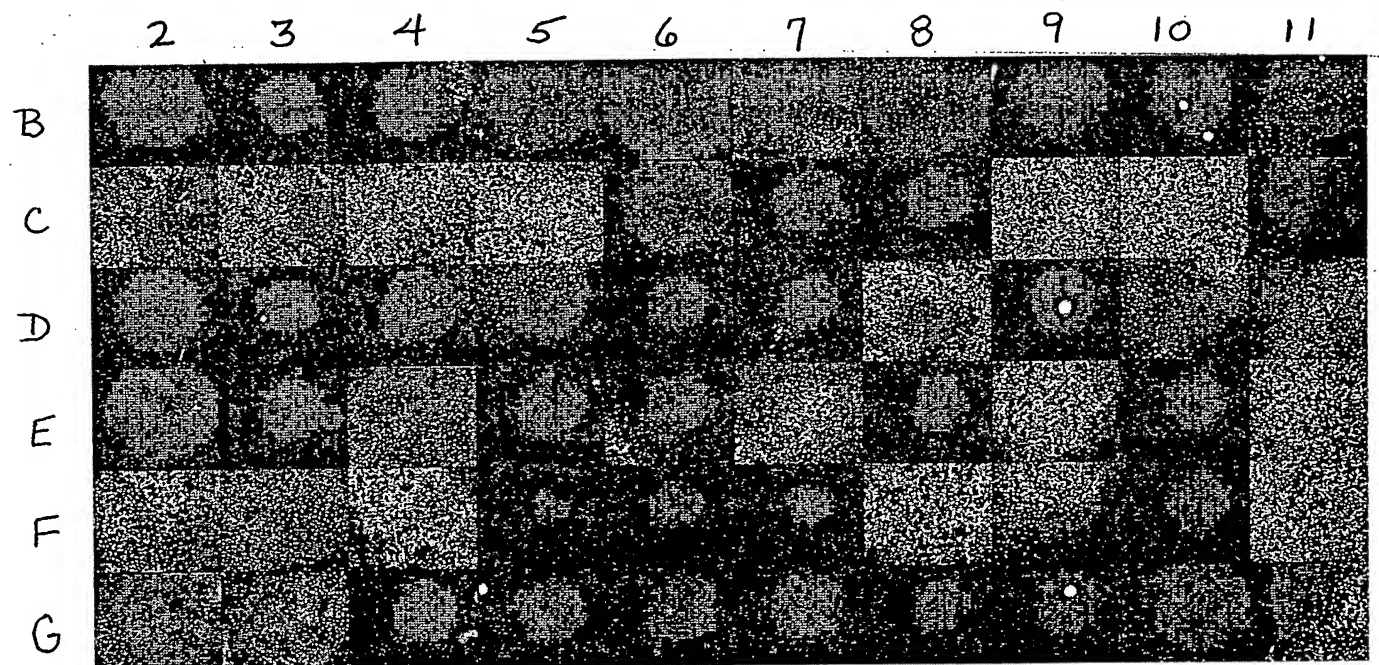
Figure 9
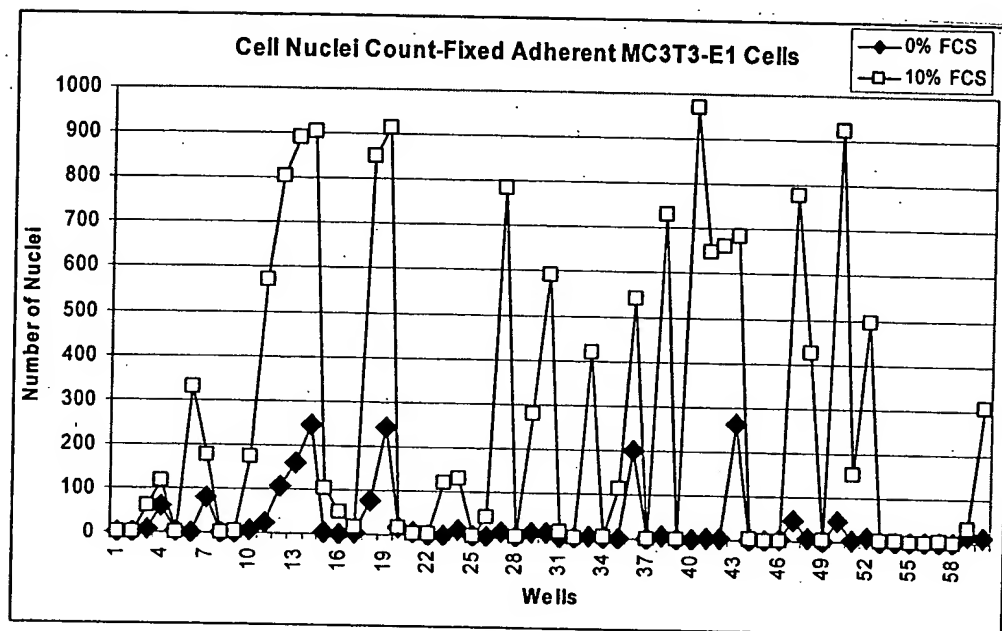
Figure 10

Cell Count - MC3T3 24hrs – No Serum

DESIGN-EXPERT Plot

Ln(Cell Count-no serum + 1.00)

Actual Components
A: Fibronectin = 5.00
B: Collagen I = 5.00
C: Vitronectin = 5.00
D: Colagen V I = 5.00
E: Collagen III = 5.00
F: Laminin = 5.00
G: Collagen IV = 5.00
H: Elastin = 5.00
J: Poly-L-Lysine = 5.00
K: Poly-L-Ornithine = 5.00

Trace (Piepel)

Deviation from Reference Blend
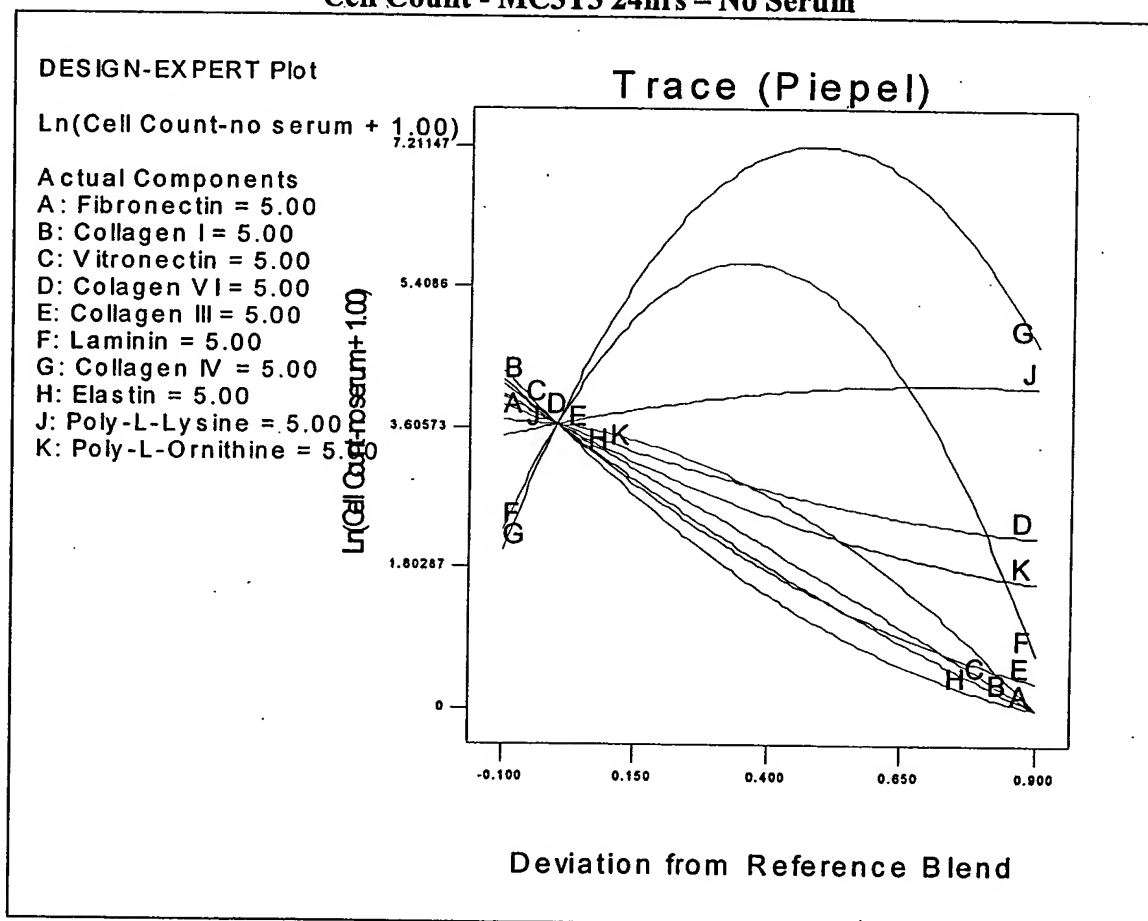
(Horizontal axis on plot is LN(Cell Count + 1)

Figure 11

Cell Count – MC3T3 24hrs - 10% Serum

DESIGN-EXPERT Plot

Ln(cell count-10% serum + 1.00)

Actual Components
A: Fibronectin = 5.00
B: Collagen I = 5.00
C: Vitronectin = 5.00
D: Colagen V I = 5.00
E: Collagen III = 5.00
F: Laminin = 5.00
G: Collagen IV = 5.00
H: Elastin = 5.00
J: Poly-L-Lysine = 5.00
K: Poly-L-Ornithine = 5.00

Trace (Piepel)

Deviation from Reference Blend
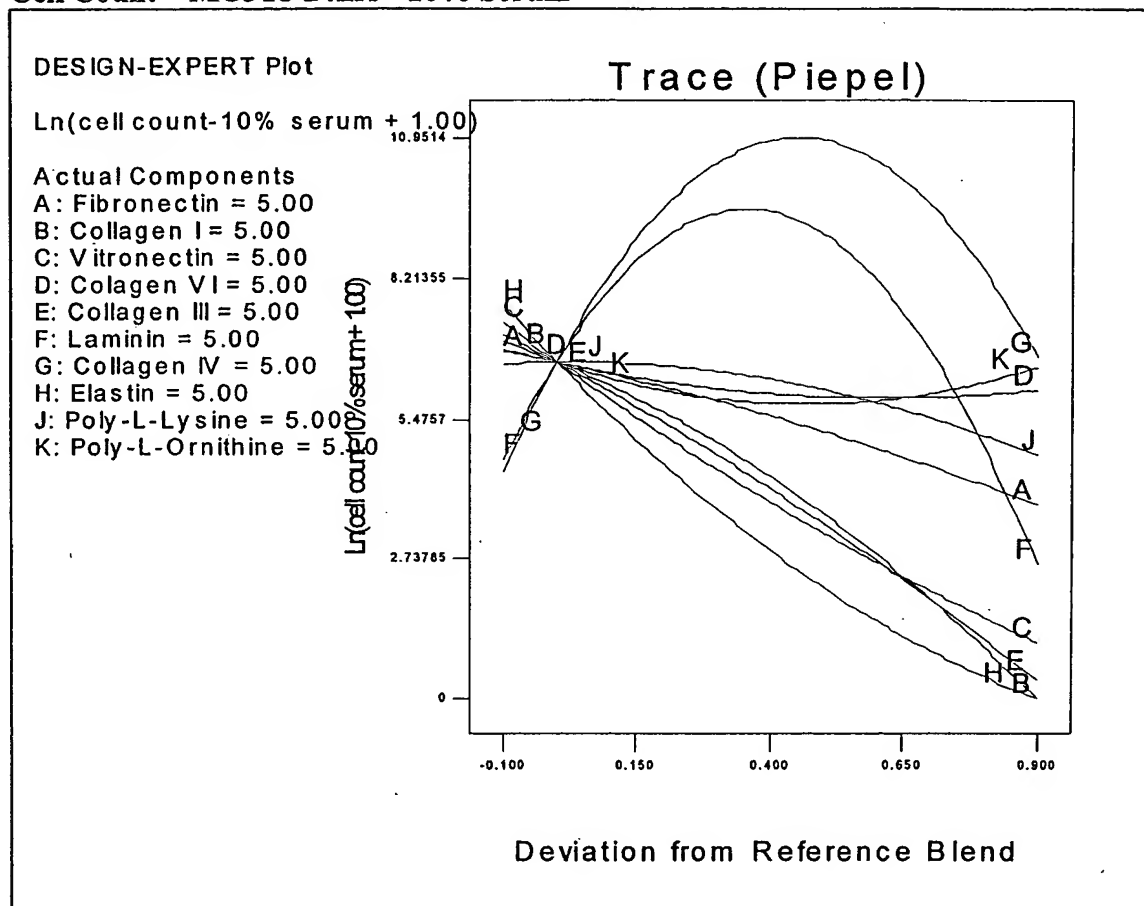
Figure 12

Run	F01	F02	F03	F04	F05	F06	F07	F08
1	-1	-1	-1	1	-1	-1	-1	1
2	-1	-1	1	1	-1	-1	-1	-1
3	1	1	1	-1	-1	-1	-1	-1
4	1	1	1	-1	1	1	1	1
5	1	-1	1	1	1	-1	1	-1
6	-1	-1	-1	1	1	-1	-1	-1
7	1	-1	1	-1	-1	-1	1	-1
8	1	-1	1	-1	1	-1	-1	1
9	-1	1	1	1	1	1	-1	-1
10	1	1	1	1	-1	-1	-1	-1
11	-1	-1	-1	-1	-1	1	1	-1
12	1	-1	-1	-1	-1	-1	1	1
13	1	1	1	-1	-1	1	1	1
14	-1	-1	-1	1	1	-1	1	1
15	1	-1	-1	1	1	1	1	1
16	-1	1	-1	1	-1	-1	-1	1
17	-1	-1	1	-1	-1	-1	1	1
18	1	-1	-1	-1	1	1	-1	1
19	1	1	-1	-1	-1	-1	1	-1
20	1	-1	-1	1	-1	1	1	1
21	-1	-1	-1	-1	1	-1	-1	-1
22	1	-1	1	-1	1	-1	-1	-1
23	-1	-1	1	-1	-1	1	1	1
24	-1	1	-1	-1	1	1	1	-1
25	-1	1	-1	1	-1	-1	1	-1
26	1	1	1	1	1	1	1	1
27	-1	1	1	1	-1	1	1	1
28	-1	1	-1	-1	-1	1	1	-1
29	1	1	-1	1	1	1	1	-1
30	1	-1	-1	1	-1	-1	1	1
31	-1	1	1	-1	1	-1	1	-1
32	1	1	-1	1	-1	1	-1	-1
33	1	1	-1	-1	-1	-1	-1	1
34	1	1	1	1	-1	-1	1	1
35	-1	-1	-1	1	-1	-1	1	-1
36	1	-1	-1	-1	-1	1	-1	-1
37	-1	1	-1	-1	1	-1	1	1
38	1	1	1	1	1	-1	-1	-1
39	1	1	-1	-1	1	1	1	1
40	-1	1	1	1	-1	1	-1	1
41	1	-1	1	1	-1	1	-1	1
42	1	1	1	-1	1	-1	1	-1
43	-1	-1	1	-1	1	1	1	-1
44	-1	1	1	1	1	-1	-1	1
45	-1	1	1	-1	1	1	-1	1
46	1	1	-1	1	1	-1	1	-1
47	1	-1	-1	-1	1	-1	-1	1
48	-1	1	-1	1	1	1	-1	1
49	-1	-1	1	1	1	1	1	-1
50	-1	-1	1	1	-1	1	1	-1
51	-1	-1	1	1	1	-1	1	1
52	1	1	-1	1	-1	1	-1	-1
53	-1	-1	-1	-1	1	1	-1	-1
54	-1	1	-1	-1	-1	1	-1	-1
55	1	-1	-1	1	1	1	-1	1
56	-1	1	-1	-1	1	-1	-1	1
57	-1	-1	1	-1	-1	1	-1	1
58	-1	1	1	-1	-1	-1	-1	1
59	1	-1	1	-1	-1	1	-1	-1
60	1	-1	1	1	1	1	-1	-1

Fig. 13a

F09	F10	F11	F12	F13	F14	F15	F16	F17
1	-1	1	1	-1	1	-1	1	-1
1	-1	-1	-1	1	1	-1	1	1
1	1	-1	-1	-1	-1	1	-1	-1
-1	-1	1	1	1	1	1	-1	-1
1	-1	-1	1	-1	-1	1	1	1
-1	1	-1	-1	-1	1	1	-1	1
-1	1	-1	1	1	1	-1	1	-1
-1	-1	1	1	1	-1	1	1	1
-1	-1	-1	1	1	-1	-1	-1	-1
-1	1	1	-1	-1	-1	-1	1	-1
-1	-1	-1	1	-1	-1	-1	1	1
-1	-1	-1	-1	1	-1	-1	-1	1
1	1	-1	-1	-1	-1	-1	1	1
-1	1	-1	1	-1	-1	-1	1	-1
-1	-1	-1	-1	-1	1	1	-1	-1
-1	-1	1	-1	1	1	1	-1	1
-1	1	1	-1	1	-1	1	-1	-1
1	-1	1	-1	1	-1	-1	-1	1
-1	-1	1	1	-1	1	1	-1	1
-1	1	-1	1	-1	-1	1	-1	-1
1	1	-1	1	1	-1	1	-1	1
1	-1	-1	1	-1	1	1	1	-1
-1	1	1	1	1	-1	-1	1	1
1	1	1	1	-1	-1	1	1	1
-1	1	1	1	-1	1	1	1	1
1	1	1	1	1	1	1	1	1
1	-1	-1	1	1	1	1	1	-1
1	1	-1	1	-1	1	-1	-1	-1
-1	1	1	1	1	1	-1	-1	-1
1	-1	1	1	1	1	-1	-1	1
-1	-1	1	-1	-1	1	-1	1	1
1	-1	-1	1	1	1	-1	1	1
1	-1	-1	-1	-1	1	-1	-1	-1
1	1	1	-1	-1	-1	-1	-1	1
1	1	1	-1	1	-1	1	-1	-1
-1	1	1	-1	1	1	-1	1	-1
1	-1	1	-1	1	-1	-1	1	-1
-1	-1	1	1	-1	-1	-1	-1	1
1	-1	-1	-1	-1	-1	1	1	-1
-1	-1	1	-1	-1	1	1	1	-1
-1	1	-1	-1	1	1	1	-1	1
1	-1	1	-1	-1	1	-1	-1	1
1	1	-1	1	-1	-1	-1	-1	-1
-1	1	-1	-1	-1	1	-1	-1	1
1	-1	-1	-1	1	-1	-1	1	-1
-1	1	1	1	-1	1	-1	1	-1
-1	1	-1	-1	1	-1	-1	1	1
-1	-1	-1	-1	1	1	-1	-1	-1
1	-1	1	-1	-1	-1	1	-1	-1
1	1	-1	-1	1	1	1	1	1
-1	1	-1	-1	1	-1	1	1	1
-1	-1	1	-1	-1	-1	1	1	-1
1	-1	1	1	1	-1	1	1	1
1	1	1	-1	-1	1	1	-1	-1
1	1	-1	1	1	1	1	-1	1
-1	-1	-1	1	1	-1	1	1	-1
1	1	1	-1	1	1	1	1	-1
1	1	1	1	1	-1	-1	-1	-1

Fig. 13b

F18	F19	F20	F21	F22	F23	F24	F25	F26
-1	-1	1	-1	-1	1	-1	1	1
-1	1	-1	1	-1	-1	-1	1	-1
-1	1	1	-1	1	1	-1	1	-1
-1	-1	-1	1	1	-1	-1	-1	-1
-1	1	1	1	1	-1	-1	1	1
1	-1	1	-1	1	-1	-1	-1	1
1	-1	-1	1	-1	-1	1	1	1
1	-1	-1	1	1	1	1	1	-1
-1	-1	-1	-1	1	1	-1	1	1
-1	-1	1	1	-1	-1	1	-1	1
-1	1	1	-1	1	-1	1	-1	-1
1	-1	1	1	-1	1	-1	1	-1
-1	-1	-1	-1	1	-1	-1	-1	1
-1	1	-1	1	1	1	-1	1	-1
-1	-1	1	-1	-1	-1	1	1	-1
-1	1	-1	-1	1	-1	-1	1	1
-1	1	-1	-1	-1	1	1	1	1
-1	-1	1	-1	1	1	1	-1	1
1	1	-1	-1	-1	1	-1	-1	1
1	1	1	-1	1	1	1	1	-1
-1	-1	-1	1	-1	-1	1	-1	1
1	-1	1	-1	-1	1	-1	-1	1
1	1	1	-1	-1	-1	-1	-1	1
1	1	-1	-1	-1	-1	-1	1	1
-1	-1	1	1	1	1	1	-1	-1
1	1	1	1	1	1	1	1	1
-1	-1	-1	-1	1	1	-1	-1	-1
-1	-1	-1	1	-1	1	1	1	-1
-1	-1	1	1	-1	-1	-1	1	1
1	1	1	1	-1	-1	-1	-1	-1
1	-1	1	-1	1	-1	-1	1	-1
1	1	-1	-1	1	1	1	-1	1
1	1	-1	1	1	-1	1	-1	1
1	-1	-1	-1	-1	1	-1	1	-1
1	-1	-1	1	1	1	-1	1	1
1	-1	-1	-1	1	-1	-1	1	-1
-1	1	1	1	-1	1	1	1	1
-1	-1	-1	1	1	-1	1	1	-1
-1	-1	-1	1	-1	-1	-1	1	1
1	1	1	1	-1	-1	1	1	1
1	1	-1	1	-1	1	-1	-1	1
1	1	1	-1	-1	1	1	1	1
1	1	-1	1	1	1	1	-1	-1
1	1	-1	-1	-1	-1	1	-1	-1
-1	1	1	1	-1	-1	-1	1	-1
1	1	1	-1	1	-1	1	-1	-1
-1	1	-1	-1	1	1	1	-1	1
1	-1	1	1	1	1	-1	-1	1
-1	1	-1	-1	-1	1	1	-1	1
1	-1	1	1	1	-1	1	-1	1
-1	-1	-1	-1	-1	1	1	-1	-1
-1	1	-1	1	-1	-1	1	-1	-1
1	1	-1	1	-1	1	-1	-1	-1
-1	-1	1	-1	-1	1	1	1	-1
1	-1	-1	-1	-1	-1	1	1	-1
1	1	1	1	1	-1	-1	-1	-1
-1	-1	1	1	1	-1	1	1	1
1	-1	1	-1	-1	-1	1	-1	-1
-1	1	1	1	1	1	-1	-1	-1
-1	1	1	-1	-1	-1	-1	1	-1

Fig. 13c

F27	F28	F29	F30
1	-1	1	-1
-1	1	-1	1
1	-1	-1	-1
1	-1	-1	-1
1	1	1	-1
-1	-1	1	-1
-1	1	1	1
-1	-1	-1	-1
-1	1	-1	1
-1	1	-1	-1
-1	1	-1	-1
-1	-1	1	-1
1	-1	1	1
1	-1	-1	1
1	1	-1	1
1	-1	1	1
-1	1	-1	1
-1	1	-1	-1
-1	1	1	1
-1	1	1	1
1	1	-1	1
1	1	-1	1
1	-1	-1	-1
-1	-1	-1	-1
-1	-1	-1	1
1	1	1	1
-1	1	-1	-1
1	-1	1	-1
-1	-1	-1	1
1	1	-1	-1
-1	1	1	1
-1	-1	-1	-1
-1	-1	-1	1
1	1	-1	1
1	1	-1	-1
1	1	1	-1
-1	-1	1	1
1	-1	1	-1
-1	1	1	-1
1	1	-1	-1
-1	-1	1	1
1	-1	-1	-1
1	1	1	1
-1	1	1	-1
-1	1	-1	-1
1	-1	-1	1
1	1	1	-1
1	1	1	1
1	-1	1	-1
-1	-1	1	-1
-1	-1	1	-1
1	1	1	-1
1	-1	-1	1
1	1	1	1
-1	-1	-1	1
-1	1	1	-1
1	-1	-1	1
1	-1	1	1
-1	-1	1	1
-1	-1	1	1

Fig. 13d

Fig. 14

MPM Factor	Factor	Receptor	Classification
F01	Sonic hedgehog amino-terminal peptide (Shh-N)	PATCHED (PTCH-1) / PTCH-2 / SMO (smoothened)	7-pass transmembrane / 7-pass transmembrane / GPCR
F02	BMP-cocktail	BMPRc-1A, BMPRc-1B, BMPRc-2	BMPR-Ser/Thr Kinase
F03	Cholesterol (water soluble formulation)	LDL Rc / SR-BI	channels & membrane transporters
F04	Leptin (human, recombinant)	Leptin Receptor	Cytokine Rc
F05	Prolactin (human, recombinant)	Prolactin Receptor	Cytokine Rc
F06	Ciliary neurotrophic factor (CNTF) (human, recombinant)	CNTFR-alpha + gp130 + LIF Rc	Cytokine Rc
F07	Amphiregulin (long form, recombinant)	EGFR	EGFR-tyrosine kinase
F08	Fibroblast Growth Factor-8c (FGF-8c) (mouse, recombinant)	FGF Rc Family	FGFR-tyrosine kinase
F09	Fibroblast Growth Factor-7 (FGF-7) = KGF	FGF Rc Family	FGFR-tyrosine kinase
F10	Vasoactive Intestinal Peptide (VIP)	VPAC1R / VPAC2R	GPCR
F11	Gastrin/CCK8-cocktail	CCK-B/Gastrin Rc	GPCR
F12	Neuropeptide Y	Neuropeptide Y Rc Family (Y1-Y6)	GPCR
F13	Thrombin/TXA2-cocktail	thromboxane A2 Receptor	GPCR
F14	C natriuretic peptide (human, porcine, rat: frag 32-53)(CNP)	Guanylate Cyclase B (GC-B) Rc (ANPR-A & ANPR-B)	Guanylyl Cyclase
F15	Interleukin-3 (IL-3) (human, recombinant)	IL3Rc-beta (aka GMCSFRc) / IL3Rc-alpha	IL-Cytokine Rc
F16	Interleukin-18 (IL-18) (human, recombinant)	IL18Rc	IL-Cytokine Rc
F17	Midkine (MK) (human, recombinant)	PTPzeta	Miscellaneous
F18	Neurturin (NTN)	GFRa1 / GFRa2 / c-ret	Miscellaneous
F19	Dibutyryl cyclic AMP	cAMP Receptor Protein Kinase (PKA)	Ser/Thr Kinase
F20	DMF (n n dimethylformamide); a polar solvent	Not receptor mediated	Small Molecule
F21	Cycloheximide (actidione)	Not receptor mediated	Small Molecule
F22	Platelet-derived endothelial cell growth factor (PD-ECGF) (aka thymidine phosphorylase)	Not Receptor mediated	Small Molecule
F23	Laminin	Laminin-Elastin Rc / alpha6 beta4 integrin	surface-matrix receptor
F24	Transforming Growth Factor beta3 (human, recombinant)	TGFBRc-1, TGFBRc-2, TGFBRc-5	TGFBR-Ser/Thr Kinase
F25	Estradiol, beta (water soluble formulation)	Estrogen Receptor-alpha (ER-A) / Estrogen Receptor-beta (ER-B) / Estrogen-related receptor alpha (ERR-A) / Estrogen-related Receptor beta (ERR-B)	Transcription Factor
F26	Hydrocortisone	Hydrocortisone Rc	Transcription Factor
F27	nuclear factor of activated T cells (NFAT) proteins (NFAT1-NFAT5)	Not Receptor mediated	Transcription Factor
F28	Hepatocyte Growth Factor (HGF, scatter factor)	c-Met (HGFR)	tyrosine kinase
F29	Growth Hormone	GH Receptor	tyrosine kinase
F30	Brain-derived Neurotrophic Factor (BDNF) (human, recombinant)	TrkB	tyrosine kinase